Evaluation of Fish-friendly Vacuum Pump Systems to Remove Salvaged Fish from Recessed Cylindrical Holding Tanks at the Tracy Fish Collection Facility

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Summary

Operations of the south Delta state and federal fish salvage facilities in California require daily collection and holding of fish, and the transport of these fish back to the Sacramento-San Joaquin River Delta, away from the facilities. These functions are of major importance for efficient return and survival of salvaged fishes; however collecting, handling, and transport methods associated with entrainment of the fishes inadvertently may cause harm that the fish salvage facilities are attempting to minimize. The Bureau of Reclamation (Reclamation) Tracy Fish Collection Facility (TFCF) consists of a system of louvers, bypasses, and collecting/holding tanks to reduce the associated fish loss of its pumping operation. The TFCF was originally designed to divert downstream migrating juvenile Chinook salmon (*Oncorhynchus tshawytscha*) from the exported flow and was not intended to divert and salvage the myriad of fish species that are entrained by the pumping practices today. High species and fish totals, along with the documented declining abundance of several fish species, and elevated quantities of vegetative debris are prompting TFCF improvements. Evaluations and improvements of both the state and federal fish salvage facilities have been ongoing for a number of years. Efforts by California Fish and Game have demonstrated problems with survival of salvaged fish after transport to the release site, and Reclamation researchers have contributed to the understanding of survivorship and injury of fishes associated with the holding tanks at TFCF (Raquel 1989, Portz 2007, Karp and Lyons 2008). These studies indicate an important need for accelerating and expanding studies at the two salvage facilities.

Fish losses due to entrainment are reduced with improved salvage operations, and the success of these operations is dependent on the survival of screened fishes. Measuring the acute physiological stress and potential direct and indirect mortality experienced by fishes during the different components of the salvage process is vital to understanding negative impacts the process may have on fish. Exposure of fishes to

stressors, such as capture and handling, can be a great concern to fisheries biologists, in that extreme or prolonged stressors may plague fish performance (*i.e.* growth, metabolism, reproduction, immune system, predator evasion) and overall health (Barton *et al.* 2002), adversely affecting population size and sustainability. Abated performance due to sublethal stresses may increase the susceptibility of these fishes to predators (indirect mortality; Olla *et al.* 1992, Mesa *et al.* 1994, Mesa *et al.* 1998).

One of the most broadly used approaches to evaluating physiological responses of fish to environmental stressors is measuring blood plasma constituents such as cortisol, lactate, and glucose (Pickering 1981, Barton and Iwama 1991, Iwama *et al.*1995). However, because some of these reflect a normal response to less extreme or prolonged stressors, from which a fish can quickly recover, an assessment of fish well being and performance should not be restricted to an examination of internal chemo-physiological changes, alone. A more complete assessment should include an examination of chemo-physiological changes, along with a measure of tissue damage, compromised performance (*e.g*., burst swimming speed), and survival over a post-treatment holding period. This combination of evaluations for a fish's well being will assess, more accurately, stress-related effects of the fish salvage process. Information will be used to identify and evaluate potential impacts of using a vacuum pump system to remove fish from the recessed, cylindrical holding tanks in times of increased salvage that is beyond the fish-safe capacity of the lift bucket to transfer fish from the holding tank to the fish holding truck.

Occasionally throughout the year dense schools of fish (*e.g.,* threadfin shad *Dorosoma petenense*) are salvaged at very high numbers and held in recessed cylindrical holding tanks awaiting transfer to the fish hauling truck. High fish densities make it difficult to transfer fish safely using the 1890-L lift bucket and as a result mortality can occur. The lift bucket fish conveyance method has been implicated as one of the greatest sources of stress for fish in the salvage process (Portz 2007) and when fish densities are high they may be transferred without water, crushing and suffocating themselves. Under these situations, a vacuum pump system may be advantageous in transferring sensitive fish species suspended in water without damage to the hauling truck. Fish vacuum pumps are widely used to unload fishing vessel catch and are used at fish hatcheries to load trucks or transfer to other ponds (Davis *et al.* 1993). While the fish vacuum pump is commonly used to transfer fish, the effects of this device on fish health and survival are poorly understood. This method may provide relief to the overburdened lift bucket removal method during high entrainment. However, the fish vacuum system may inadvertently cause physiological stress and damage because of pressures exerted on the fish and external physical damage from working parts that potentially can lead to direct and indirect mortality. An evaluation of a commercially available fish vacuum pump system should be undertaken at Reclamation's Denver Hydraulics Laboratory and if deemed to be "fish friendly" implemented at the TFCF.

Problem Statement

South Delta fish salvage facilities serve a major importance for maintaining the survival of salvaged fishes, however occasional high fish entrainment numbers can be detrimental to fish. High fish densities make it difficult to transfer fish safely using the 1890-L lift bucket and as a result mortality can occur. Under these situations, a vacuum

pump system may be advantageous in transferring sensitive fish species suspended in water without damage to the hauling truck, relieving some of the lift bucket burden. However, the fish vacuum system may inadvertently cause physiological stress and damage because of pressures exerted on the fish and external physical damage from working parts that potentially can lead to direct and indirect mortality. An evaluation of a commercially available fish vacuum pump system should be undertaken at Reclamation's Denver Hydraulics Laboratory and if successful implemented at the TFCF in 2010.

Goals and Hypotheses

Goals:

1. Determine if fish vacuum pump systems add to the acute physiological stresses experienced by juvenile Chinook salmon.

2. Determine if fish vacuum pump systems affect scale loss and external tissue damage in juvenile Chinook salmon.

3. Determine if fish vacuum pump systems affect the burst swimming performance of juvenile Chinook salmon, possibly hindering their ability to avoid predator capture

4. Determine if fish vacuum pump systems affect the short-term survival (96 h) of juvenile Chinook salmon.

Hypotheses:

1. If a fish vacuum pump system is physiological stressful to juvenile Chinook salmon, then this transfer method should have heightened plasma cortisol, glucose, and lactate concentrations compared to a control group and/or salmon transferred with a 1890-L lift bucket.

2. If a fish vacuum pump system affect scale loss and external tissue damage in juvenile Chinook salmon, then fish experiencing this transfer technique will have greater areas of skin ulcerations and damage compared to control fish.

3. If a fish vacuum pump system affects the burst swimming performance of juvenile Chinook salmon, then maximum swimming velocities of fish experiencing this transfer technique will be slower and maximum C-start angles higher (less bending) compared to control fish.

4. If a fish vacuum pump system affects the short-term survival (96 h) of juvenile Chinook salmon, then fish experiencing this transfer technique will have greater mortality compared to control fish.

Materials and Methods

Source and Care of Fish

Sacramento River Chinook salmon used in this study will be obtained in March 2010 from the Mokelumne River Hatchery (Clements, California) or the Coleman

National Fish Hatchery (Anderson, California), and transported to the Denver Federal Center (Denver, Colorado). Juvenile fall-run Chinook salmon will be maintained in 757-L circular tanks equipped with aerated, recirculated dechlorinated municipal water. Fish will be held under a natural photoperiod (39° 43' N latitude) with natural and halogen light, and fed Silver Cup salmon feed pellets (Nelson and Son, Inc., Murray, Utah) at 1.5–2% body weight per day. Treatment and control salmon may be marked with implanted, colored microspheres on dorsal and anal fins with a high pressure needle (Photonic tagging; New West Technology, Arcata, California) to differentiate various treatment fish so they can be consolidated for a 96-h survival holding period.

The Experiment: Effects of Holding Tanks and Conveyance Methods

The experiment will be organized to evaluate the physiological stress response, scale loss and external tissue damage, swimming performance, and short-term survival (96 h) of juvenile Chinook salmon (*ca*. 110 mm) of transferring juvenile salmon vertically 6 m using a fish vacuum pump system from a holding tank into a 757-L tank simulating the fish conveyance process to the fish transport trucks. The fish vacuum pump system will be assessed to determine its affects on fish well being and performance. Twenty treatment fish will be released in the simulated recessed, cylindrical holding tank and transferred via a fish vacuum pump system to an external 757-L tank. Water quality (***i.e.,*** temperature, dissolved oxygen concentration, pH) will be monitored throughout the study. The minimum of 12 replicates will be collected testing the vacuum pump. Additionally, we will make recommendations from our finding to transfer fish from the holding and deliver them in the best possible condition to the transport truck (***e.g.***, water-to-water transfer, minimize plunging into lift bucket).

Physiological Stress Response

A control will be captured and removed from previously undisturbed 757-L tanks with modified 10-cm by 18-cm dip nets with a 1.5-L plastic reservoir sewn into the cod-end, so that fish could be transferred in water to minimize stress. All transfers of control fish will be accomplished quickly (<30 s) with minimal disturbance and handling trauma to the fish. Twenty treatment fish will be nettled, placed in a 18.9-L bucket, and inserted in the simulated recessed, cylindrical holding tank where they will be transferred vertically 6m using a fish vacuum pump system. Control and treatment fish after recapture will be quickly transferred to a bath containing a lethal dose of tricaine methanesulfate (MS-222, Argent Chemical Laboratories, Inc., Redmond, Washington; 200 mg/L), which immobilizes them in less than 30 s. This anesthetic dose inhibits stress-related increases in plasma cortisol concentration in salmon. Blood will be collected from the severed caudal peduncle in 40-µl, heparinized microhematocrit capillary tubes. Blood samples from the treatment groups under the two holding conditions will be collected at 0-, 1-, 4-, 8-, and 24-h post-treatment. Weights (±0.01 g) and measurements (TL, ±1 mm) of each fish using an electronic balance and fish measuring board will be recorded. Collected blood will be immediately centrifuged using a microhematocrit centrifuge (Clay-Adams Autocrit Ultra3) for 4 min at 12,000 x g to separate the plasma from the packed cells (Becton Dickinson Diagnostics, Sparks, Maryland). Hematocrit (packed cell volume) will be measured shortly after collection. Plasma obtained with from each fish will be transferred into plastic cryogenic freezing

vials and temporarily stored in a 10-L liquid-nitrogen dewar flask (-196 ˚C). These samples will then be shipped to Denver, Colorado, were they will be stored in a -80 ºC freezer for storage for analyses of plasma cortisol, lactate, and glucose once field component is complete. Plasma cortisol concentrations will be measured using a modified enzyme immunoassay (ELISA) at the University of California, Davis Endocrinology Lab, and plasma lactate and glucose will be measured with a polarographic analyzer (YSI 2700 Select, Yellow Springs Inc., Yellow Springs, Ohio) in the Fisheries and Wildlife Group's Fish Physiology Lab.

External Tissue Damage

Scale loss and external tissue damage will be determined in the control and the treatment group immediately post-treatment and after a 96-h holding period in 190-L tanks using fluorescein (AK-Fluor®, Akorn, Inc., Decatur, Illinois). Fluorescein is a nontoxic fluorescent dye that can be used to rapidly and easily detect scale loss and tissue lesions and ulcers by binding to breaks or tears in the epithelial barrier of soft tissue. Fish will be anesthetized in a MS-222 bath (40 mg/L) and transferred to a solution of 0.20-mg fluorescein/1-ml water for 5 min and then rinsed in three separate clean water baths for 2 min. The fish will then be euthanized in a 200-mg/L MS-222 bath and immediately examined for skin damage under an ultraviolet light (Model UVGL-58, Mineralight, Upland, California). Photographs are taken in complete darkness under ultraviolet light using a Nikon D-100 digital camera. Severity of tissue damage will be categorized, external bacterial infections will be diagnosed, and total damaged area will be quantified. Weights (±0.01 g) and measurements (TL, ±1 mm) of each fish using an electronic balance and fish measuring board will be recorded.

Burst Swimming Performance

After being transferred using a fish vacuum pump system, a juvenile salmon will be quickly recaptured and delivered to an acrylic raceway for measuring burst swimming performance (including mean velocity, maximum velocity, mean acceleration, maximum acceleration, and C-start angles). Burst swimming performance will be determined for control directly out of culture tanks and treatment fish immediately after recapture from holding tank scenarios. The burst swimming raceway (220-cm length with a 30-cm-wide swimming channel) will be filled to 25-cm depth to minimize vertical swimming. Startle responses and burst swimming speeds will be filmed with a Phantom v4.2 high-speed camera (Vision Research, Inc., Wayne, New Jersey) fitted with a wide angle lens and lighted by four, 150-W floodlights situated 1.3 m above the raceway. Fish will be stimulated to swim with a tethered tennis ball that strikes the water directly behind the fish. The high-speed camera system will record fish burst swimming motions as it swims to the opposite end of the raceway, at 500 frames/s. The high-speed video recordings will be analyzed image-by-image (Peak Performance Technologies, Inc., Centennial, Colorado) to determine velocities and acceleration rates at specific distances, and fast-start body orientation (C-shape). Maximum burst swimming velocity will be determined as the greatest distance moved over a specified elapsed time (cm/s). Acceleration will be calculated as increasing velocity up to maximum burst swimming speed (m/s^2). The software automatically calibrates the pixels/cm with the filming information (resolution, recording speed), and the fish can then be tracked by two points on a centimeter grid. For

determining C-start angles, we will compare a video segment before the C-start preparation stage, where the fish is mostly straight, to when it contracts and bends into a "C" shape to establish three points to measure contraction angles. Angle theta (θ) will be determined to be the angle made from the two intersecting lines meeting at the center of mass. Theta (θ) is recorded as the minimum angle when <180º and as the minimum explementary angle when >180º. Using the equation 0.35+ (0.2TL), where TL is the total length (mm) of a salmonid to determine the center of mass, we will manually track the trailing edge of the caudal fin, head, and center of mass points for each fish image-by-image. Weights (±0.01 g) and measurements (TL, ±1 mm) will be recorded for each fish using an electronic balance and measuring board.

168-Hour Survival Monitoring

Survival will be determined over a 96-h holding in 190-L tanks with the tanks examined daily for mortalities. Mortalities are removed daily so water quality is not degradated. After 96 h, surviving fish will be counted, weighed (±0.01 g), and measured (TL, ±1 mm) using an electronic balance and fish measuring board.

Data Analyses

Statistical analyses will be performed using Sigmastat 3.0 (Jandel Scientific, San Rafael, California) software package. Differences between the treatments and control will be tested using a either a t-test or analysis of variance (ANOVA; Zar 1984, Steel *et al.* 1997). The Tukey's test will be used for all pair-wise multiple comparisons for parametric data. The Shapiro-Wilk's test for normality and the Levene's test for homogeneity of variances will be used to determine ANOVA assumptions. Data that do not meet the ANOVA assumptions and are unable to be power or log transformed will be compared with a Kruskal-Wallis non-parametric ANOVA on ranks with the Dunn's test for pairwise multiple comparisons (Zar 1984, Steel *et al.* 1997). Differences will be considered significant at $P < 0.05$.

Coordination and Collaboration

This research will be a collaborative effort between Fisheries and Wildlife Research Group staff and TFCF biologists. Research will be coordinated directly with the Tracy Technical Advisory Team, Tracy Fish Facility Improvement Program manager and the Tracy Fish Collection staff. Opportunities for participation of state and federal resource agencies in research-related discussions at the Tracy Technical Review Team and Central Valley Fish Facilities Review Team meetings will be offered. A draft Tracy Series Report for will be made available in the fall of 2010.

Endangered Species Concerns

This study will not involve the use of wild endangered or threatened species. Chinook salmon will be obtained from the Mokelumne River Hatchery (Clements, California) or Coleman National Fish Hatchery (Anderson, California). The laboratory evaluation of a fish vacuum pump system will not impact listed species.

Dissemination of Results (Deliverables and Outcomes)

The primary deliverable will be articles published in both the Tracy Volume Series and a peer-reviewed scientific journal. Technical updates will also be provided to the Tracy Technical Advisory Team and the Central Valley Fish Facilities Review Team, along with posters and oral presentations given at scientific forums. Additionally, information gained on the successes and limitations of the fish collection and salvage process will help guide future improvements in the fish collection, holding, and transport process.

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